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Original Article

Kisspeptin and body weight homeostasis in relation to phenotypic features of polycystic ovary syndrome; metabolic regulation of reproduction

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ABSTRACT

Background: Polycystic ovary syndrome (PCOS) is a heterogeneous disorder characterized by a diverse collection of reproductive and metabolic abnormalities. kisspeptin (KISS) is novel peptides associated with regulation of metabolism, food intake, puberty and reproduction. **The aim** of the present study was to estimate KISS level in patients with PCOS, and to evaluate the possible relationship between KISS level with anthropometric measures as well as clinic-morphological features of PCOS.

Materials and methods: cross section control study enrolled 90 control group and 105 patients with PCOS and they were stratified according to their body mass index (BMI) to; underweight (n = 9, BMI ˂19), normal weight (n = 25, BMI = 19.1–25), over weight (n = 34,BMI = 25.1–30), obese grade I (n = 12, BMI = 30.1–35) , obese grade II (n = 13, BMI 35.1–40) and obese grade III (n = 12, BMI˃40).Circulating KISS levels were measured using ELISA.

Results: Our results revealed that, KISS levels were higher in PCOS patients compared to controls. Among PCOS group, there were significant lower level of KISS levels in underweight, overweight and obese compared to normal weight group. Even more importantly, KISS levels decreased with increasing of BMI as the following, grade I, grade II and grade III. Moreover, it was negatively correlated to anthropometric measures, glycemic, lipid profile and positively correlated the phenotype characteristics of PCOS. Linear regression test observed that hirsutism score, HOMA-IR and LH were the main predictors of KISS levels in PCOS.

Conclusion: circulating KISS is an important regulator of body weight and reproduction especially in PCOS women.

1. Introduction

The unfolding global malnutrition epidemic apparently reflects powerful interactions between genes and our changing environment. Today, more than thirty percent of population globally suffers from at least one form of malnutrition: wasting, stunting, vitamin and mineral deficiency, overweight and obesity [1].

Extensive data support a major role of weight homeostasis in reproduction approximately 1–5% of women suffer from 'weight-related amenorrhea.Apart from delayed puberty which is relatively common in underweight girls [2], obesity also is associated with polycystic ovary syndrome (PCOS) which is a common endocrinopathy in women during reproductive age [3] and accounts for approximately 75% of anovulatory infertility disorders [4].

Several lines of evidence indicate the pathophysiological mechanisms underpinning PCOS which are multifactorial, including genetic and metabolic factors [5].A preponderance of evidence suggests that PCOS is associated with an increase in LH pulse amplitude and pulse frequency [6].

Kisspeptins (KISS) are peptide products of the KISS1 gene.There are various forms of KISS which in turn regulate the reproductive axis by stimulation of their cognate receptor KISS1R on gonadotropin releasing hormone (GnRH) neurons, thereby regulating luteinizing hormone (LH) and follicle stimulating hormone (FSH) secretion [7]. Compelling evidence suggests that KISS expressed in

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both central [8] and peripheral tissues including metabolic tissues like fat, liver, and pancreas [9].

A growing body of evidence has corroborated that appetite and the reproductive axis are closely linked to nutritional status and disturbances in positive and negative energy balance often result in impairments of fertility. Therefore, we aimed in the current study to estimate KISS level in patients with PCOS, and to evaluate the possible relationship between KISS level with anthropometric measures as well as clinic-morphological features of PCOS.

2. Material and methods

A cross-sectional controlled study was planned on 105 women with PCOS. The enrolled PCOS women classified according to body mass index (BMI) to; underweight (n = 9, BMI ˂19), normal weight (n = 25, BMI = 19.1–25), over weight (n = 34,BMI = 25.1–30), obese grade I (n = 12, BMI = 30.1–35) , obese grade II (n = 13, BMI 35.1–40) and obese grade III (n = 12, BMI˃40) in addition to and 90 aged and BMI matched healthy fertile control women. The diagnosis of PCOS was based on the 2004 revised Rotterdam criteria [10], with at least two of the following features: (i) oligo-ovulation or chronic anovulation, (ii) clinical and/or biochemical hyperandrogenism, and (iii) ultrasound appearance of polycystic ovaries. Other etiologies that could mimic PCOS were excluded by doing appropriate blood tests depending on the clinical suspicion - included a history of hyperandrogenic states (such as non-classical congenital adrenal hyperplasia, androgen secreting tumors, Cushing's syndrome, 21-hydroxylase deficiency, or hyperprolactinemia), DM, hypertension, liver, kidney, or thyroid diseases. Moreover, women with steroid or oral contraceptive drug intake in the preceding 3 months as well as previously diagnosed diabetes were also excluded from the study.

Oligo-ovulation and/or anovulation was characterized by oligomenorrhea (intermenstrual intervals of ≥35 days) and amenorrhea (intervals >3 months). Clinical hyperandrogenism was defined as the presence of hirsutism (Ferriman-Gallwey score of ≥8) and/or acne. Polycystic ovary on ultrasound was defined as the presence of at least one ovary 10 cm^3 or more in volume and/or at least one ovary with 12 or more follicles measuring 2–9 mm in diameter. All patients were subjected to thorough history taking and full clinical assessment including blood pressure. Height, waist circumference (WC) and hip circumference (HC) were measured to calculate obesity indices. Anthropometric variables including Body mass index (BMI) was calculated as weight in kg/height in (meters) 2, waist-to-hip ratio (WHR), waist circumference (cm)/hip circumference (cm), mid upper arm circumference (MAC) taken midway between the olecranon process of the ulna and the acromion process of the scapula in cm.Body compositions including fat mass (FM) and fat free mass (FFM) were measured by Dual-energy X-ray absorptiometry (DEXA). The ethical committee of Faculties of Medicine, Zagazig University approved.

2.1. Sampling of blood and biochemical analysis

The blood samples of all study's subjects were drawn after an overnight fast and divided into 3 portions: 1 ml of whole blood was collected into EDTA tubes, for HbA1c; 1 ml of whole blood was collected into potassium oxalate and sodium fluoride containing tubes for fasting plasma glucose (FPG). Sera were separated from remaining sample part and stored at −20 °C until analysis. Total cholesterol (TC) and triglycerides (TG) levels were measured by routine enzymatic methods (Spinreact, Girona, Spain). HDL cholesterol concentration was determined after precipitation of the apoB-containing lipoproteins. LDL cholesterol level was calculated using the Friedewald formula.We measured, fasting serum insulin (FSI), FSH, LH, total testosterone, we calculated insulin resistance (IR) with the homeostatic model assessment-IR (HOMA-IR) index, which is defined as FSI value (lU/mL) × FPG value (mg/dl)/405. The β-cell function was calculated using HOMA-β as follows: {20*[FSI (μU/mL)]/[FPG (mmol/L) - 3.5]}. KISS levels were measured with an enzyme-linked immunoassay kit (ELISA kit, Phoenix Pharmaceuticals Inc., Belmond, CA), after extraction with Phoenix Peptide sepcolumns (RK-Sepcol-2).

2.2. Statistical analysis

Statistical analyses were performed using the Statistical Package for the Social Sciences for Windows (version 21.0; SPSS Inc., Chicago, IL, USA). Data were expressed using descriptive statistic (mean ± standard deviation) and were analyzed using “t” test. One-way analysis of variance (ANOVA) test was done to compare different parameters between more than two groups. Pearson correlation coefficient was used to assess the association between. A linear regression analysis was performed to detect the main predictors of KISS concentration in women with PCOS. Receiver operating characteristic (ROC) analysis was performed to assess the potential accuracy of plasma KISS for diagnosis of PCOS, the area under the curve (AUC), and the cutoff values. We considered P to be significant at <0.05.

3. Results

3.1. General characteristics of the subjects

General characteristics of the study subjects are summarized in Table 1. Our results showed in age and BMI matched case and control women, as expected there were significantly higher values of PCOS phenotypic features in particularly; hirsutism score, ovarian volume, AFC, FSH, LH, LH/FSH, DHEA-S, androstenedione, and total testosterone. Moreover, PCOS women had significantly higher values of TC, TG, and LDL cholesterol, FPG, FSI, HbA1c and HOMA-IR. On the contrary, patients with PCOS had significantly lower levels of HDL and HOMA −β, when compared with controls, p ˂0.001.

3.2. General characteristics of controls and PCOS women stratified by BMI

Obesity plays an important role in the pathogenesis of PCOS as according to our cross-section study, the majority of PCOS patients were overweight or obese (67.7%) we stratified PCOS patients by BMI.In underweight group (n = 9), there were significantly higher values of DHEA-S compared to normal weight group (n = 250).On the other hand, LDL level was significantly lower in underweight group compared to normal weight group, p ˂0.001.Considering overweight group (n = 34), there were significantly higher values of TG, FSI and DHEA-S compared to normal weight group. While, there was significantly lower values of HDL in overweight group compared to normal weight group. In obese group (n = 37), there were significantly higher values of AFC, TC, TG, LDL, FSI,HOMA-IR,DHEA-S and androstenedione. On the other hand, HDL level was significantly lower in obese group compared to normal weight group, p ˂0.001, (Table 2).

3.3. Comparison of KISS (pg|mL) levels in studied groups

PCOS patients (0.31 ± 0.1) had significantly higher levels compared to control group (0.27 ± 0.06) (Fig. 1A). Among PCOS group, there were significant differences between underweight (0.21 ± 0.09), overweight (0.36 ± 0.09) and obese (0.29 ± 0.065),

Table 1
Clinical, anthropometric and laboratory characteristics of studied groups.

	Control group (mean ± SD) (n = 90)	PCO patients (mean ± SD) (n = 105)	P
Age (years)	32.44 ± 7.08	30.6 ± 6.57	0.062
Systolic blood pressure (mm Hg)	127.73 ± 6.51	127.7 ± 6.5	0.076
Diastolic blood pressure (mm Hg)	85.2 ± 3.84	86.5 ± 4.4	0.025
Hirsutism score	5.44 ± 0.746	8.86 ± 4.36	˂0.001*
BMI (kg/m2)	27.73 ± 6.37	28.4 ± 7.872	0.500
Waist/hip ratio	1.16 ± 0.193	1.23 ± 0.264	0.342
MAC (cm)	25.2 ± 6.52	26.3 ± 8.67	0.302
Weight (kg)	69.1 ± 15.97	71.09 ± 19.68	0.444
FM(kg)	22.4 ± 5.09	22.75 ± 6.29	0.686
FMI(Kg/m2)	6.24 ± 1.49	6.25 ± 1.731	0.946
FFM(kg)	46.4 ± 11.23	48.3 ± 13.3	0.292
FFMI (kg/m2)	21.78 ± 5.646	22.1 ± 6.14	0.638
Ovarian volume	5.17 ± 0.88	7.85 ± 3.84	˂0.001*
AFC	6.29 ± 1.39	8.68 ± 4.29	˂0.001*
Total cholesterol (mg/dL)	166.6 ± 20.6	195.65 ± 17.3	˂0.001*
Triglycerides (mg/dL)	179.4 ± 11.27	302.6 ± 85.15	˂0.001*
LDL cholesterol (mg/dL)	105.9 ± 4.36	137.9 ± 17.429	˂0.001*
HDL cholesterol (mg/dL)	41.26 ± 4.30	34.7 ± 4.736	˂0.001*
FPG (mg/dL)	85.1 ± 8.97	93.7 ± 6.71	˂0.001*
FSI (lU/mL)	4.8 ± 0.97	5.57 ± 1.35	˂0.001*
HOMA-IR	1.42 ± 0.33	3.31 ± 2.0	˂0.001*
HOMA-β	146.5 ± 60.7	105.15 ± 99.5	˂0.001*
HbA1c (%)	5.7 ± 0.144	5.9 ± 0.116	˂0.001*
Total testosterone (ng/mL)	0.5 ± 0.14	0.78 ± 0.22	˂0.001*
FSH (mIU/mL)	4.8 ± 0.977	5.57 ± 1.35	˂0.001*
LH (mIU/mL)	6.5 ± 1.20	7.1 ± 1.28	˂0.001*
LH/FSH	1.52 ± 0.38	1.34 ± 0.36	˂0.001*
DHEA-S (mg/mL)	0.93 ± 0.51	1.26 ± 0.814	˂0.001*
Androstenedione (ng/mL)	1.2 ± 0.35	1.8 ± 0.53	˂0.001*

BMI; body mass index, MAC; mid arm circumferences, FM; fat mass, FMI; fat mass index, FFM; fat free mass,FFMI; fat free mass index, FSI; fasting serum insulin, FPG; fasting plasma glucose,AFC; antral follicle cells, HOMA-IR; homeostasis model assessments of insulin resistance, DHEA; dehydroepiandrosterone . *P < 0.05 when compared with control group.

Table 2
Clinical, anthropometric and laboratory characteristics of PCOS groups.

	PCO patients (n = 105)			Obese (mean ± SD) (n = 37)
	Under weight (mean ± SD) (n = 9)	Normal weight (mean ± SD) (n = 25)	Over weight (mean ± SD) (n = 34)	
SBP (mm Hg)	133.8 ± 4.83	131.3 ± 6.36	130.7 ± 7.49	126.2 ± 8.45
DBP (mm Hg)	86.01 ± 4.47	87.4 ± 4.13	87.4 ± 4.25	85.27 ± 4.55
Hirsutism score	6.56 ± 1.36	7.44 ± 1.33	9.39 ± 5.14	10.36 ± 4.71
Body mass index (kg/m2)	17.1 ± 1.05	21.56 ± 1.63	26.47 ± 1.05	37.6 ± 4.498
Waist/hip ratio	0.691 ± 0.2	0.83 ± 0.20	0.84 ± 0.20	0.87 ± 0.32
MAC (cm)	14.1 ± 1.05	18.56 ± 1.63	25.4 ± 4.52	35.54 ± 5.41
Weight (kg)	42.7 ± 2.63	53.9 ± 4.0	66.17 ± 2.628	94.1 ± 11.2
Ovarian volume	6.4 ± 2.05	7.8 ± 4.63	8.88 ± 4.1	7.88 ± 1.7
AFC	7.2 ± 1.39	6.9 ± 1.71	8.73 ± 5.0	10.31 ± 4.63[c]
FM(kg)	13.68 ± 0.84	17.24 ± 1.3	21.17 ± .84	30.11 ± 3.59
FMI(Kg/m2)	3.76 ± 0.23	4.74 ± 0.35	5.82 ± 0.231	8.28 ± 0.98
FFM(kg)	29.08 ± 1.79	36.6 ± 2.77	36.6 ± 2.779	64.01 ± 7.6
FFMI (kg/m2)	13.3 ± 0.82	16.8 ± 1.2	20.6 ± 0.819	29.3 ± 3.5
Total cholesterol (mg/dL)	193.2 ± 15.13	201.9 ± 9.67	200.2 ± 9.9	188.1 ± 23.5[c]
Triglycerides (mg/dL)	252.1 ± 93.6	252.8 ± 12.8	310.4 ± 61.4[b]	340.4 ± 61.4[c]
LDL cholesterol (mg/dL)	124.2 ± 12.2[a]	147.19 ± 4.37	147.4 ± 7.21	126.8 ± 21.1[c]
HDL cholesterol (mg/dL)	36.2 ± 4.73	37.6 ± 6.14	32.6 ± 2.19[b]	32.5 ± 1.19[c]
FPG (mg/dL)	94.2 ± 3.80	95.2 ± 5.67	94. ±6.71	91.8 ± 7.66
FSI (lU/mL)	9.24 ± 5.5[a]	13.6 ± 4.645	18.6 ± 10.43[b]	24.8 ± 4.88[c]
HOMA-IR	2.1 ± 1.281	3.18 ± 1.611	4.39 ± 2.501	5.76 ± 1.01[c]
HOMA-β	115.2 ± 86.5	105.1 ± 76.9	97.25 ± 28.9	89.01 ± 44.1
HbA1c (%)	5.8744 .1	5.95 ± 0.11	5.99 ± 0.05	5.91 ± 0.14
Total testosterone (ng/mL)	0.69 ± 0.2	0.83 ± 0.2	.84 ± 0.2	.87 ± 0.32
FSH (mIU/mL)	5.75 ± 1.46	5.49 ± 1.42	5.8 ± 1.42	5.313 ± 1.21
LH (mIU/mL)	6.96 ± 1.57	7.99 ± 1.89	7.1 ± 0.516	7.03 ± 1.105
LH/FSH	1.36 ± 0.378	1.37 ± .346	1.42 ± 0.43	1.27 ± 0.35
DHEA-S (mg/mL)	1.5 ± 0.845[a]	0.7 ± 0.505b	1.17 ± 0.736[b]	2.09 ± 0.769[c]
Androstenedione (ng/mL)	1.87 ± 0.51	1.98 ± 0.48	1.45 ± 0.46	2.06 ± 0.443[c]

BMI; body mass index, MAC; mid arm circumferences, FM; fat mass, FMI; fat mass index, FFM; fat free mass,FFMI; fat free mass index, FSI; fasting serum insulin, FPG; fasting plasma glucose,AFC; antral follicle cells, HOMA-IR; homeostasis model assessments of insulin resistance, DHEA; dehydroepiandrosterone; [a] significant difference between underweight vs normal weight, [b]significant difference between overweight vs normal weight significant difference between obese vs normal weight.

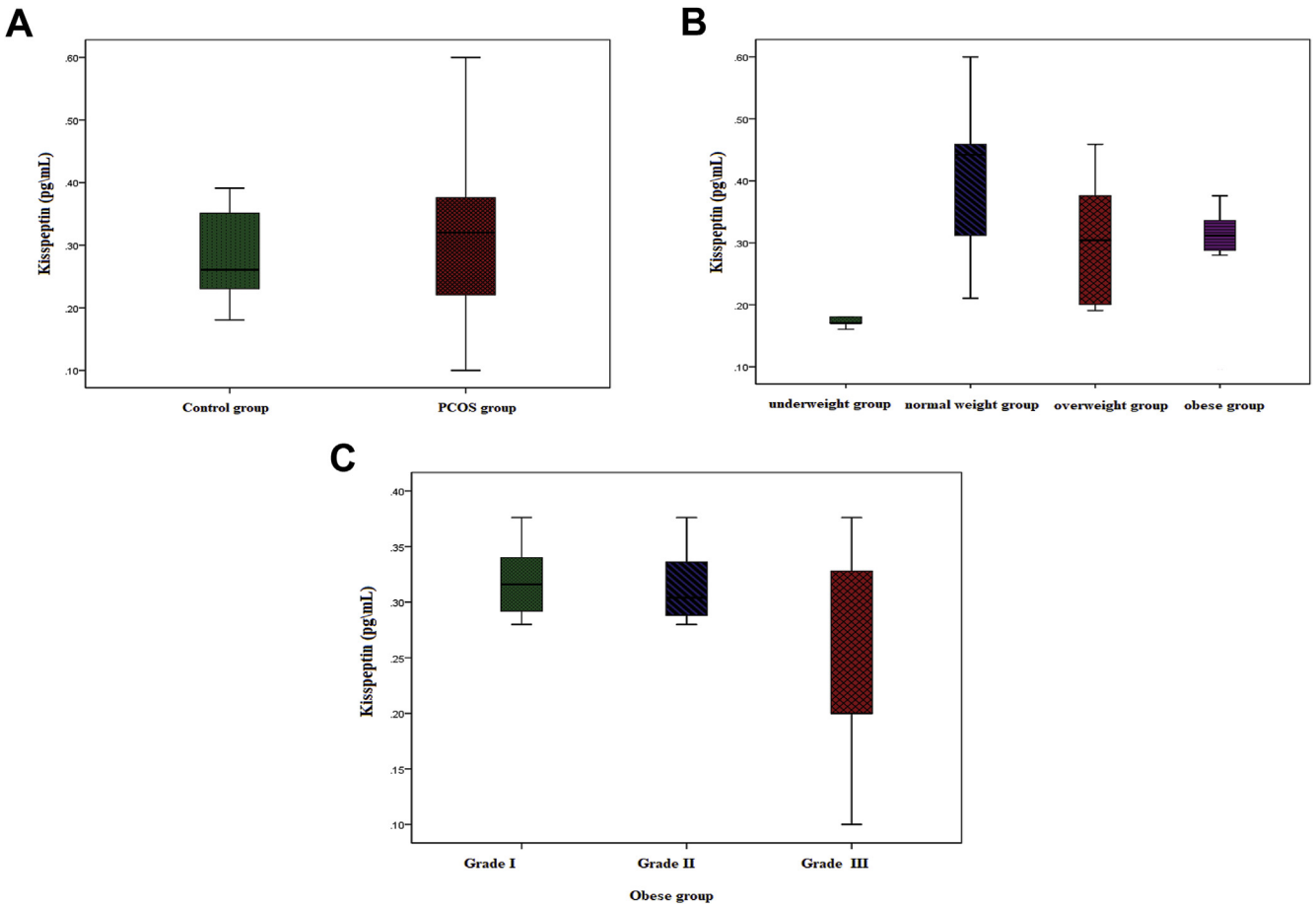

Fig. 1. A): Kisspeptin (pg\mL) levels in the studied groups. 1B): Kisspeptin (pg\mL) levels in PCOS groups. 1C): Kisspeptin (pg\mL) levels in the obese PCOS groups.

compared to normal weight group (0.38 ± 0.106) (Fig. 1B). In order to better educate the association between severity of obesity and KISS (pg\mL) levels, we classified obese group to, grade I (BMI = 30.1–35), grade II (BMI 35.1–40) and grade III (BMI˃40).we found the level of KISS (pg\mL) decreased with increasing of BMI as the following, grade I (0.31 ± 0.031), grade II (0.30 ± 0.053) and grade III (0.23 ± 0.09), p ˂0.001 (Fig. 1C),

3.4. Correlation between KISS (pg|mL) with clinical and biochemical parameters of PCOS patients

In PCOS group, ($n = 120$), KISS levels were significantly negative correlated with FMI% and MAC. Furthermore, KISS levels were significantly positive correlated with PCOS phenotypic features; hirsutism score, ovarian volume, AFC, LH, total testosterone and androstenedione. Even more interestingly, KISS levels were negatively correlated with cardio metabolic factors; TG, LDL FSI and HOMA-IR, p ˂0.001 (Table 3).

3.5. Linear regression analysis with KISS (pg|mL) levels as dependent variable in PCOS groups

In PCOS group, linear regression analysis revealed that only hirsutism score, HOMA-IR and LH were the main predictors of KISS levels among other clinical and laboratory biomarkers of PCOS, $p < 0.05$ (Table 4).

3.6. Accuracy of KISS (pg|mL) for diagnosis of PCOS by ROC analysis

The power of *KISS (pg|mL)* levels to diagnose PCOS among studied subjects was evaluated using ROC analysis. The AUC was 0.592 (95% CI = 0.510–0.674 with sensitivity = 64.8%, specificity = 69.9%, and the cutoff values (0.2844). When compared our patients with control, (Fig. 2).

4. Discussion

PCOS is one of the most common causes of infertility due to anovulation; the clinical features of PCOS are heterogeneous and may change throughout the lifespan, starting from adolescence to postmenopausal age [11]. The high prevalence of obesity in women with PCOS has profound effects on both the pathophysiology and the phenotypic features of PCOS [12].

KISS is peptide involved in regulation of body weight, metabolism and sexual functions. Changes in energy status or metabolic signals affect both reproduction and hypothalamic KISS levels, which in turn mediate metabolic effects on reproductive status [13,14]. We aimed in the current study to estimate KISS level in patients with PCOS, and to evaluate the possible relationship between KISS level with anthropometric measures as well as clinic-morphological features of PCOS.

In line with our previous studies, the present data provide compelling evidence that there were statistically significant elevations of cardio metabolic risk factors in PCOS compared to controls [15–19].

Table 3
Pearson correlation of Kisspeptin (pg\mL) with clinical, anthropometric and phenotypic features of PCOS group.

Characteristics	Kisspeptin	
	r	p
Hirsutism score	0.279	˂0.001*
Body mass index (kg/m2)	0.182	0.063
Waist/hip ratio	0.178	0.070
MAC (cm)	−0.342	˂0.001*
Weight (kg)	0.181	0.081
Ovarian volume	0.340	˂0.001*
AFC	0.268	˂0.001*
FM(kg)	−0.315	˂0.001*
FMI(Kg/m2)	0.171	0.072
FFM(kg)	0.164	0.071
FFMI (kg/m2)	0.153	0.081
Total cholesterol (mg/dL)	0.055	0.579
Triglycerides (mg/dL)	0.297	˂0.001*
LDL cholesterol (mg/dL)	0.368	˂0.001*
HDL cholesterol (mg/dL)	0.058	0.554
FPG (mg/dL)	0.121	0.218
FSI (lU/mL)	0.199	˂0.001*
HOMA-IR	0.206	˂0.001*
HOMA-β	−0.023	0.814
HbA1c (%)	0.171	0.081
Total testosterone (ng/mL)	0.268	˂0.001*
FSH (mIU/mL)	0.045	0.649
LH (mIU/mL)	0.267	˂0.001*
LH/FSH	0.125	0.203
DHEA-S (mg/mL)	0.034	0.730
Androstenedione (ng/mL)	0.278	˂0.001*

FSI, fasting serum insulin; FPG, fasting plasma glucose; AFC, antral follicle cells; FMI, fat mass index; FFMI, fat free mass index; HOMA-IR, homeostasis model assessments of insulin resistance; DHEA-S, dehydroepiandrosteron sulfate. *P < 0.05.

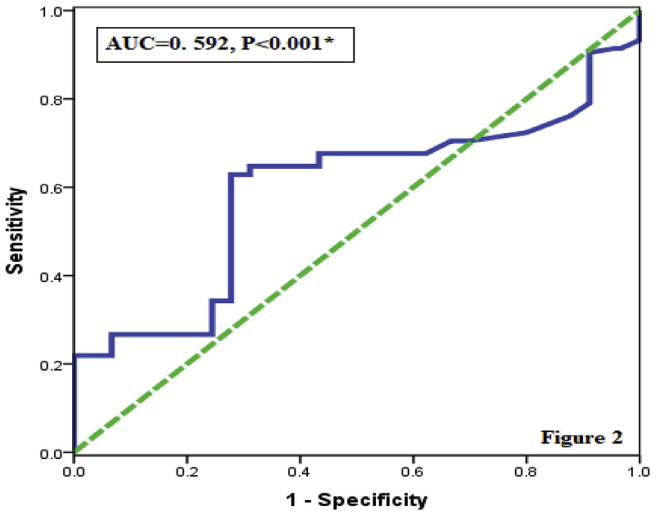

Fig. 2. Accuracy Kisspeptin (pg\mL) for discriminating PCOS from control group by ROC analysis.

The interesting finding of the present study is that, in PCOS group, there was a significant higher values of KISS compared to control group with matched age and BMI.

Similar to our result, a study conducted by Gorkem et al. observed that women with PCOS had increased serum KISS levels compared to controls [20].

In agreement with our results, Yilmaz et al. observed metastin (KISS) levels were higher in women with PCOS as compared to controls regardless of BMI [21].

Our findings are in concordance with Katulski et al. who detected a significantly higher KISS pulse frequency as well as integrated serum concentrations group of PCOS patients with oligomenorrhea than the eumenorrheic group of PCOS subjects [22].

Considering KISS level in PCOS among other Arabic population, a study conducted on Saudi women found that women with PCOS had higher KISS levels compared to controls, but the values were non-significant. This controversy could be due to significantly higher BMI in PCOS group compared controls [23].

Conflicting data have been reported for KISS levels in various cohorts of PCOS women.A study conducted by. Jeon et al. reported higher KISS levels in women with PCOS [24]. While another study reported by Yerlikaya et al. observed lower levels in women with PCOS as compared to controls they suggested this different finding may be due to obesity and insulin resistance in their studied women that may have negative impact on kiss levels [25].

There are intriguing reports suggesting that KISS had a key role in metabolism regulation in the peripheral tissues [26] as its receptor are expressed in peripheral tissues controlling metabolism [27] and metabolic diseases; undernutrition, obesity, and diabetes were associated with dysregulation of KISS levels [28]. Experimental studies confirmed the metabolic roles of KISS in peripheral tissues, lipid metabolism [29] In addition to obesity and insulin resistance [30].

The results presented here are innovative; as this study was the first Egyptian study that investigated the possible association of KISS level and body weight homeostasis, as we sub classified our PCOS group to four group according to BMI to assess the level of KISS in underweight PCOS women as well as obese PCOS women. Even more importantly, we subdivided obese PCOS women according to severity of obesity for accurate and better evaluation of KISS levels in different groups. We found that among PCOS group, there were significant lower level in underweight, overweight and obese compared to normal weight group. In order to better educate the association between severity of obesity and KISS (pg\mL) levels, we classified obese group to three groups according to BMI, grade I

Table 4
Linear regression analyses in PCOS women to test the influence of the main independent variables against Kisspeptin (pg\mL) (dependent variable) in PCOS women.

Model		Unstandardized Coefficients		Standardized Coefficients	t	P value	95% C.I.	
		B	SE	Beta			Lower Bound	Upper Bound
1	(Constant)	119.092	23.515		5.065	0.000	69.243	168.941
	MAC (cm)	0.186	0.494	0.070	0.377	0.711	−0.861	1.234
	Ovarian volume	0.142	0.663	−0.066	−0.214	0.833	−1.548	1.265
	AFC	0.125	0.645	−0.049	−0.193	0.849	−1.492	1.243
	Hirsutism score	1.702	0.644	0.532	2.642	˂0.05*	0.336	3.069
	FPG	0.055	0.148	0−.071	−0.371	0.715	−0.368	0.258
	HOMA-IR	1.197	0.408	−0.684	−2.934	˂0.01*	−2.061	−0.332
	Total cholesterol	0.069	0.074	0.153	0.938	0.362	−0.087	0.226
	LH	3.104	1.424	0.385	2.180	˂0.05*	0.085	6.122

(BMI = 30.1–35), grade II (BMI 35.1–40) and grade III (BMI>40).we found the level of KISS (pg\mL) decreased with increasing of BMI as the following, grade I, grade II and grade III.

A study conducted by Chen et al. revealed that plasma KISS were increased in lean adolescent and adult women with PCOS compared with lean adolescent control group. Also, other study observed lower KISS levels in overweight or obese PCOS women [31,32].

We in this study attempted to pierce out the association between KISS levels and anthropometric measures as well as phenotypic features of PCOS. Our results found KISS levels were significantly negative correlated with FMI% and MAC. Even more interestingly, KISS levels were negatively correlated with cardio metabolic factors; TG, LDL FSI and HOMA-IR.

Our data are in line with previous findings reporting by Tolson et al. they observed that KISS levels were associated with reproduction, adiposity, metabolism, and glucose homeostasis, especially in adult females [32].

In agreement with our results, Kołodziejski et al. explored the negative associations between KISS levels and obesity dyslipidemia and insulin resistance [32].

Considering the associations between KISS levels and PCOS phenotypic features, our findings confirmed the significantly positive correlated with PCOS phenotypic features; hirsutism score, ovarian volume, AFC, LH, total testosterone and androstenedione.

Similar to our results, an interesting study from Gorkem et al. suggested that KISS concentrations were negatively correlated with serum FSH and positively correlated with serum T. testosterone and DHEAS levels [20].

In conclusion we found that KISS levels were higher in PCOS patients compared to controls. Among PCOS group, there were significant lower level of KISS levels in underweight, overweight and obese compared to normal weight group. Even more importantly, KISS levels decreased with increasing of BMI as the following, grade I, grade II and grade III. Moreover, it was negatively correlated to anthropometric measures, glycemic, lipid profile and positively correlated the phenotype characteristics of PCOS.

Conflicts of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.dsx.2019.04.017.

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